Assessment of salt treatment and effect of citric acid as salt stress alleviator on soybean (*Glycine max L.*)

[1]Deepak Choudhary, [2]Angurbala Bafna, [3]Tasneem Rangwala, [4]Swati Mali
[1]Research Scholar, [2]Professor and Head, [3]Assistant Professor, [4] Research Scholar
[1,2,3,4]Department of Biochemistry,
[1,2,3,4] Govt. Holkar (Autonomous, Model) Science College, Indore
[1]bnp100deepak@xyz.com, [2]abbafna@gmail.com,
[3]tasneem.rangwala0303@xyz.com,[4]swatimali282@gmail.com

***Abstract*— Salinity stress is one of the major abiotic factors limiting crop productivity worldwide, especially in legumes such as soybean (*Glycine max* L.), which are highly sensitive to elevated salt concentrations. The present investigation was undertaken to evaluate the ameliorative potential of citric acid, an organic acid and a key intermediate of the tricarboxylic acid (TCA) cycle, in mitigating the detrimental effects of salt stress on soybean. Varying concentrations of NaCl (50, 100, 150 and 200 mM) were given to plants two times at the interval of 15 days. Growth and stress parameters were analyzed and recorded. Results revealed that citric acid application significantly improved height of plants, weight and length of pods in comparison to plants grown under salt stress alone. Biochemical analyses showed a marked decrease in lipid peroxidation and an increase in antioxidant enzyme peroxidase activity upon citric acid treatment. These findings indicate that citric acid plays an important role in alleviating salinity-induced oxidative stress, maintaining cellular redox homeostasis, and improving plant performance under saline environments.**

***Index Terms*— NaCl toxicity, Length of pods, MDA content, Peroxidase enzyme, Weight of Pods.**

I. Introduction

Salinity is a major environmental constraint affecting the productivity and sustainability of agricultural systems. More than 20% of irrigated land worldwide is affected by salinization, leading to reduced crop yield and soil fertility. In saline conditions, excess sodium chloride (NaCl) disrupts ionic balance, induces osmotic stress, and generates reactive oxygen species (ROS), causing damage to proteins, lipids, and nucleic acids [1].

Soybean (*Glycine max* L.) is a nutritionally important leguminous crop valued for its high protein and oil content. However, it is highly susceptible to salinity, particularly during early stages of growth. Salt stress leads to chlorosis, reduced leaf expansion, and inhibition of photosynthetic efficiency due to reactive oxygen species in soybean plants. Therefore, exploring eco-friendly strategies to enhance salt tolerance is crucial [2].

Citric acid (CA), a low-molecular-weight organic acid and a key metabolite in the Krebs cycle, plays a significant role in energy metabolism, ion chelation, and stress signaling [3]. Exogenous application of citric acid has been reported to improve plant growth under various abiotic stresses, including drought and salinity, by modulating antioxidant defense systems and stabilizing cellular membranes. The present study aimed to investigate the effect of foliar application of citric acid on growth and oxidative stress parameters of soybean under NaCl-induced salinity stress.

II. Material and method

Location of the study - Study was conducted in open field at Loharpipliya village, Dewas road (M.P). Geographical location of fields selected as per latitude longitude read by GPS 76 Garmin was N- 22 54’40”-41.4” E- 075 59” 35.1”- 37”. Wheat seeds were sown in square blocks of 2*2 meter2. Randomized block design was used for treatment. Each block contains approx. 80-100 plants. Following treatments were given in respective blocks. Samples were analyzed for oxidative stress parameters, at the Department of Biochemistry, Govt. Holkar (Autonomous, Model) Science College, Indore.

- ***Collection of seeds*** - Seeds of soybean were collected from Beej Bhandar.
- ***Variety of seeds*** - NRC 157
- ***Time interval of the study*** - from June 2021 to October

Experimental design –

- ***Salt treatments*** - Varying concentrations of NaCl (50, 100, 150 and 200 mM) were given to plants two times at the interval of 15 days. First treatment was given on 5th day of sowing seeds and second treatment 20th day of sowing seed.
- ***Foliar application of citric acid –*** Citric acid solution of concentration 0.32g/L was applied as foliar spray after 15 days of 2nd salt treatment i.e. 35th day of sowing seeds and second - after 30 days 2nd salt treatment i.e. 50th day of sowing seeds.

- ***Total 9 treatments were designed for Soybean as follows:***

Table II.1 Treatment design

S. No.	Codes	Treatments
1.	**T1**	Control (irrigated with only water)
2.	**T2**	50 mM
3.	**T2CA**	50 mM + 0.32g/L citric acid
4.	**T3**	100 mM
5.	**T3CA**	100 mM + 0.32g/L citric acid
6.	**T4**	150 mM
7.	**T4CA**	150 mM + 0.32g/L citric acid
8.	**T5**	200 mM
9.	**T5CA**	200 mM +0.32g/L citric acid

Parameters studied –

1. ***Growth parameters***
   - ***Height of plant –***

The height soybean plants were measured three times, i.e. after 15 days of 2nd salt stress, after 15 days of 1st foliar spray and after 15 days of 2nd foliar spray, using standard centimeter scale. Measurements were taken in triplicates.

- ***Weight of Pods of soybean –***

The weight of 10 pods of soybean was measured twice, i.e. after 15 days of 1st foliar spray and after 15 days of 2nd foliar spray, using a digital balance. Before weighing, each pod was cleaned to remove any soil or moisture, then gently dried with a paper towel. weight of 10 pods of soybean was recorded immediately to avoid any weight change due to moisture loss. Measurements were taken in triplicates.

- ***Length of Pods of soybean –***

The length of each pod was measured twice, i.e. after 15 days of 1st foliar spray and after 15 days of 2nd foliar spray, using a standard centimeter scale and then converted to mm. The measurement was taken from the base of the pod to the tip. While measuring the length of pods of soybean a care was taken to ensure that the pods were laid flat to avoid curvature errors. Measurements were taken in triplicates. Measurements were taken in triplicates.

2. ***Oxidative stress parameters*** –
   - ***MDA Content –***

The malondialdehyde (MDA) content was measured in the leaves of soybean and wheat after 15 days of the second salt stress, the first foliar spray, and the second foliar spray, as well as in the grains after harvest. MDA, an indicator of lipid peroxidation [4], was estimated following Heath and Packer (1968). A 100 mg tissue sample was homogenized in 1 ml of 10% TCA under cold conditions, and 1 ml of the homogenate was mixed with 4 ml of TBA reagent. The mixture was heated at 95°C for 30 min, cooled, and centrifuged at 1900 rpm for 10 min. The absorbance of the supernatant was recorded at 532 nm using a visible spectrophotometer, with a blank prepared using 10% TCA. MDA concentration was calculated using the formula: - ($A = \varepsilon \cdot l \cdot c$) with $\varepsilon = 155\ mM^{-1}\ cm^{-1}$.

- ***Peroxidase enzyme activity –***

Peroxidase activity was measured following Summer (1943). Fresh seed tissue was homogenized in 3 mL of ice-cold 0.1 M potassium phosphate buffer (pH 6.0) and centrifuged at 16,000 × g for 20 min at 4°C. The supernatant served as the enzyme source. Activity was determined by the oxidation of o-dianisidine in the presence of H_2O_2, forming a yellow-orange product measured at 430 nm [5]. One unit of enzyme activity was defined as the amount causing an increase of 1.0 in absorbance per minute under assay conditions.

III. RESULTS

Height of plants–

After 15 days of the second salt stress, plant height declined in all treatments compared to T1, with the greatest reduction observed in T5CA. Following the first foliar spray, plant height decreased significantly across all treatments relative to T1, with T5CA showing the highest reduction, indicating that the first foliar spray had no significant ameliorative effect on plant height. However, after the second foliar spray, a significant increase in plant height was observed in T2CA and T5CA compared to their respective salt-stressed controls (T2 and T5), suggesting the beneficial effect of the second foliar application.

Table III.1 Effect of 2nd salt treatment and foliar spray of citric acid

Treatments	Height of plants (cm)		
	After 15 days of 2nd salt stress	*After 15 days of 1st foliar spray*	*After 15 days of 2nd foliar spray*
T1	34.33±1.15	58.33±1.53	76.33±1.53
T2	28.67±2.31^{**a} (-16.50)	53.67±3.21^{**a} (-8.00)	74.33±0.58^{**a} (-2.62)
T2CA	31.67±1.53^{NS} (-7.77)	47.33±1.15^{*a} (-18.86)	90.67±3.51$^{**a\ ***b}$ (18.78)
T3	31.33±1.15^{*a} (-8.74)	47±2.65^{***a} (-19.43)	70.33±0.58^{***a} (-7.86)
T3CA	30±2^{*a} (-12.62)	43.67±0.58^{***a} (-25.14)	83.67±4.04$^{*a\ *c}$ (9.61)
T4	31.67±1.53^{NS} (-7.77)	43±1^{***a} (-26.29)	74±3.46^{NS} (-3.06)
T4CA	30.67±0.58^{**a} (-10.68)	41.67±0.58^{***a} (-28.57)	82.33±0.58$^{**a\ *d}$ (7.86)
T5	29.33±1.53^{**a} (-14.56)	39.67±0.58^{***a} (-32.00)	68.33±6.35^{NS} (-10.48)
T5CA	26.67±1.53^{**a} (-22.33)	38.33±1.15^{***a} (-34.29)	77.67±3.21^{NS} (1.75)

a, b, c, d, and e indicate p value relative to T1, T2, T3, T4, T5 treatment respectively. NS indicates insignificant difference i.e. p value > 0.05, * indicate p value < 0.05 (significant difference), ** indicate p value < 0.01 (highly significant difference), *** indicate p value < 0.001 (extremely significant difference). Parenthesis indicate percent increase or decrease.

Weight of pods –

Fifteen days after the first foliar spray, pod weight increased significantly in T4CA compared to T4, while no significant changes were observed in T5CA relative to T5. Following the second foliar spray, pod weight increased significantly in T5CA compared to T5, and in T2CA relative to T2, demonstrating that the second foliar spray enhanced pod development under salt stress.

Pod length –

Fifteen days after the first foliar spray, pod length increased significantly in T4CA compared to T1, T3CA as compared to T3, T5CA as compared to T5, while fifteen days after second foliar spray, pod length increased significantly in T2CA as compared to T2, and in T5CA as compared to T5.

Table III.2 Effect of 1st and 2nd foliar spray of citric acid on weight and length of pods

Treatments	Weight of 10 Pods (gm)		Length of Pods (mm)	
	After 15 days of 1st foliar spray	*After 15 days of 2nd foliar spray*	*After 15 days of 1st foliar spray*	*After 15 days of 2nd foliar spray*
T1	3.047±0.311	3.597±0.295	20.667±1.528	40.367±1.026
T2	3.01±0.18^{NS} (-1.20)	3.18±0.519^{NS} (-11.59)	20.033±0.95^{NS} (-3.06)	49.12±1.025^{NS} (21.68)
T2CA	3.493±0.766^{NS} (14.66)	3.617±0.655^{NS} (0.56)	22.167±1.258^{NS} (7.26)	41.667±1.258^{*b} (3.22)
T3	2.903±0.286^{NS} (-4.70)	2.963±1.052^{NS} (-17.63)	19.067±0.902^{NS} (-7.74)	39.333±1.607^{NS} (-2.56)
T3CA	3.487±0.343^{NS} (14.44)	3.56±0.684^{NS} (-1.03)	22.167±0.577^{**c} (7.26)	40.833±1.041^{NS} (1.15)
T4	2.56±0.036^{*a} (-15.97)	2.727±0.064^{**a} (-24.19)	18.033±1.05^{NS} (-12.74)	39.433±1.102^{NS} (-2.31)
T4CA	2.813±0.114^{*d} (-7.66)	3.06±0.94^{NS} (-14.93)	16.5±0.866^{**a} (-20.16)	30.667±1.155^{NS} (-24.03)
T5	2.05±0.225^{**a} (-32.71)	2.467±0.54^{*a} (-31.42)	17.767±1.25^{NS} (-14.03)	30.167±0.289^{NS} (-25.27)
T5CA	2.477±0.447^{NS} (-18.71)	3.873±0.552^{*e} (7.67)	20.367±0.808^{*e} (-1.45)	31.667±0.577^{*e} (-21.55)

a, b, c, d, and e indicate p value relative to T1, T2, T3, T4, T5 treatment respectively. NS indicates insignificant difference i.e. p value > 0.05, * indicate p value < 0.05 (significant difference), ** indicate p value < 0.01 (highly significant difference), *** indicate p value < 0.001 (extremely significant difference). Parenthesis indicate percent increase or decrease.

MDA content –

Fifteen days after the second salt stress, MDA levels increased markedly in all treatments compared to T1, reflecting enhanced oxidative stress, with the highest increase in T5CA. After the first foliar spray, MDA content decreased significantly in T4CA compared to T4, and in T3CA compared to T3, indicating a partial alleviation of oxidative stress. No significant changes in MDA were observed following the second foliar spray in the selected treatments.

Table III.3 Effect of 2nd salt treatment and foliar spray of citric acid on MDA content

MDA content			
Treatments	***After 15 days of 2nd salt stress (in mM/g)***	***After 15 days of 1st foliar spray (in μM/g)***	***After 15 days of 2nd foliar spray (in μM/g)***
T1	0.002±0	0.048±0.001	0.056±0.047
T2	0.004±0.001^{**a} (140)	0.058±0.008^{NS} (21.00)	0.065±0.02^{NS} (16.07)
T2CA	0.007±0$^{***a\ **b}$ (280)	0.065±0.003^{**a} (35.23)	0.057±0.051^{NS} (1.79)
T3	0.008±0.001^{***a} (350)	0.098±0.005^{**a} (102.85)	0.067±0.015^{NS} (19.64)
T3CA	0.008±0^{***a} (390)	0.064±0.003$^{**a\ ***c}$ (31.67)	0.07±0.011^{NS} (25.00)
T4	0.009±0.001^{***a} (440)	0.042±0.002^{**a} (-12.10)	0.092±0.015^{NS} (64.29)
T4CA	0.01±0^{***a} (480)	0.06±0.002$^{**a\ ***d}$ (23.13)	0.078±0.027^{NS} (39.29)
T5	0.011±0.001^{**a} (560)	0.072±0.011^{NS} (49.11)	0.072±0.022^{NS} (28.57)
T5CA	0.013±0.001^{***a} (680)	0.096±0.001^{***a} (99.29)	0.088±0.022^{NS} (57.14)

a, b, c, d, and e indicate p value relative to T1, T2, T3, T4, T5 treatment respectively. NS indicates insignificant difference i.e. p value > 0.05, * indicate p value < 0.05 (significant difference), ** indicate p value < 0.01 (highly significant difference), *** indicate p value < 0.001 (extremely significant difference). Parenthesis indicate percent increase or decrease.

Peroxidase enzyme activity –

After the second salt stress, POD activity increased significantly in all treatments compared to T1, indicating activation of the antioxidant defense system, with the highest increase observed in T5CA. Following the first foliar spray, POD activity increased significantly in T2CA and T4CA compared to T2 and T4, respectively, while T3CA showed a decrease compared to T3. After the second foliar spray, a significant decrease in POD activity was observed in T2CA compared to T2, suggesting modulation of enzymatic activity by the second foliar application.

Table III.4 Effect of 2nd salt treatment and foliar spray of citric acid on peroxidase enzyme activity

Peroxidase enzyme activity (units/min/g)			
Treatments	***After 15 days of 2nd salt stress***	***After 15 days of 1st foliar spray***	***After 15 days of 2nd foliar spray***
T1	0.002±0	5.625±1.25	13.292±6.124
T2	0.004±0.001^{**a} (140)	7.5±1.25^{NS} (33.33)	9.458±0.813^{NS} (-28.84)
T2CA	0.007±0$^{***a\ **b}$ (280)	12.708±0.191$^{**a\ **b}$ (125.93)	11.167±1.377^{NS} (-15.99)
T3	0.008±0.001^{***a} (350)	9.292±0.591^{*a} (65.19)	6.933±2.159^{NS} (-47.84)
T3CA	0.008±0^{***a} (390)	4.583±0.617^{***c} (-18.52)	11.433±0.43^{NS} (-13.99)
T4	0.009±0.001^{***a} (440)	8.083±0.564^{*a} (43.70)	8.117±1.696^{NS} (-38.93)
T4CA	0.01±0^{***a} (480)	8.067±0.494^{NS} (43.41)	8.704±1.182^{NS} (-34.52)
T5	0.011±0.001^{**a} (560)	4.35±0.054^{NS} (-22.67)	10.908±2.597^{NS} (-17.94)
T5CA	0.013±0.001^{***a} (680)	10.625±0.451$^{**a\ ***e}$ (88.89)	8.313±3.558^{NS} (-37.46)

a, b, c, d, and e indicate p value relative to T1, T2, T3, T4, T5 treatment respectively. NS indicates insignificant difference i.e. p value > 0.05, * indicate p value < 0.05 (significant difference), ** indicate p value < 0.01 (highly significant difference), *** indicate p value < 0.001 (extremely significant difference). Parenthesis indicate percent increase or decrease.

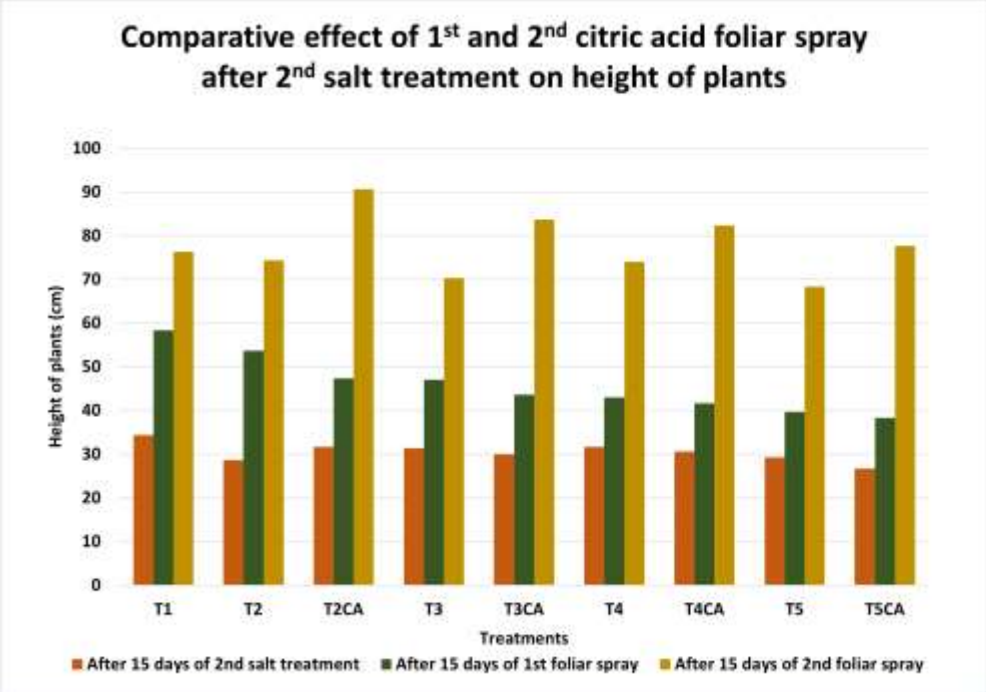

Figure III.1Effect of citric acid foliar spray after 2nd salt treatment on height of plants

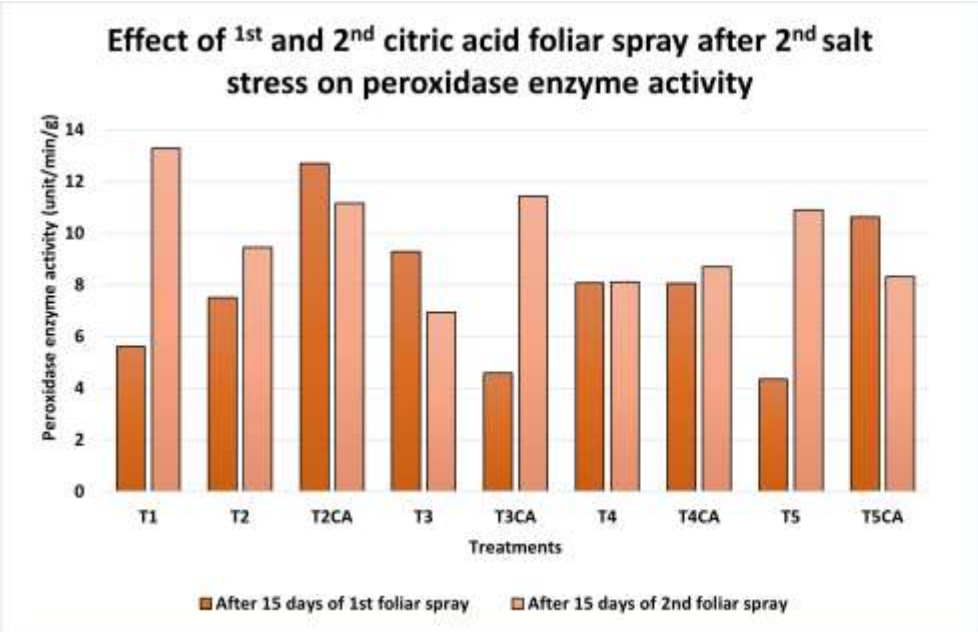

Figure III.2 Effect of citric acid foliar spray after 2nd salt stress on peroxidase activity

IV. DISCUSSION

The findings are consistent with previous research, that increase in salinity level causes decrease in plant height and number of leaves. Numerous studies have reported that higher NaCl concentrations lead to a significant decrease in plant height [6],[7]. The previous study conducted by [8], reported a significant reduction in plant growth under salt treatment, likely due to osmotic stress and ion toxicity that interfere with water uptake, nutrient balance, and cell elongation. The finding also supports the previous study [9] that the CA topical application affected various growth parameters including plant height in salt stressed as well as non-salt stressed control plants. salt treatment causes decrease in weight and length of pods of okra, due to high concentrations of sodium chloride ions and these ions cause decrease in cell growth, damage the cell membrane and can even alter the physiological functions of plants like respiration and photosynthesis that ultimately causes reduced growth [10], [11], [12]. CA exogenous application improved weight and length of pod of Vigna mungo in study [3], and suggests that CA neutralizes the sodium chloride toxic effects in plants. A previous study also found the significant effect of silica application on weight and length of pod [10].

MDA is widely recognized as a marker for oxidative stress, originating from lipid peroxidation processes that occur during cell membrane damage [13]. The current study results are in Accordance to a previous study that the treatments with 100mM NaCl did not significantly alter MDA levels compared to untreated plants, indicating that mild salinity stress may not immediately trigger lipid peroxidation [14]. A similar trend was observed by another study, where no significant rise in MDA was detected under 70mM NaCl, although prolonged exposer to 140 mM NaCl for 30 days resulted in noticeable increase [15].

Peroxidase (POD) enzyme plays critical roles in scavenging hydrogen peroxide by converting it into water and oxygen, thus protecting cells from oxidative damage [16], [17]. If peroxidase enzyme activity decreases with the increasing salt concentration, suggesting a weaker antioxidant response and possibly lower salinity tolerance, whereas, if POD enzyme activity increases with increasing salt concentration. Suggesting a higher antioxidant response and showing higher salt tolerance [18]. Application of silicon has been proposed an alternative strategy to alleviate salinity in crops [19].

V. Conclusion

The study demonstrated that citric acid effectively alleviates salt stress in soybean (*Glycine max* L.). Foliar application of citric acid significantly improved growth traits such as plant height, pod length, and pod weight under salinity. It reduced MDA content, indicating lower lipid peroxidation and better membrane stability, while enhancing peroxidase activity and antioxidant defense. Hence, citric acid plays a vital role in improving stress tolerance and overall physiological performance of soybean plants grown under saline environments.

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References

[1] Acosta-Motos, J. R., Ortuño, M. F., Bernal-Vicente, A., Diaz-Vivancos, P., Sanchez-Blanco, M. J., & Hernandez, J. A. (2017). Plant responses to salt stress: adaptive mechanisms. *Agronomy*, *7*(1), 18.

[2] Phang, T. H., Shao, G., & Lam, H. M. (2008). Salt tolerance in soybean. *Journal of integrative plant biology*, *50*(10), 1196-1212.

[3] Hossain, Md. A., Khatun, Mst. S., Hosen, M., Sayed, Z. I., Islam, Md. R., Chowdhury, Md. K., Iqbal, M. A., Al-Ashkar, I., Erden, Z., Toprak, C. C., Sabagh, A. El, & Islam, M. S. (2024). Citric Acid Alleviated Salt Stress by Modulating Photosynthetic Pigments, Plant Water Status, Yield and Nutritional Quality of Black gram [Vigna mungo (L.) Hepper]. *LEGUME RESEARCH - AN INTERNATIONAL JOURNAL*, *Of*. https://doi.org/10.18805/LRF-820

[4] Heath R. L. and Packer L. (1968). Photoperoxidation in isolated chloroplasts. I. Kinetics and stoichiometry of fatty acid peroxidation. Archives in Biochemistry and Biophysics. 125: 189–198.

[5] Summer J. B., Gjessing E. C. (1943). Arch Biochem., 2, 291.

[6] Gama, P. B. S., Inanaga, S., Tanaka, K., & Nakazawa, R. (2017). Physiological response of common bean (Phaseolus vulgaris L.) seedlings to salinity stress. *African Journal of Biotechnology*, *6*(2), 079–088.

[7] Mustard, J., & Renault, S. (2006). Response of red-osier dogwood (*Cornus sericea*) seedlings to NaCl during the onset of bud break. *Canadian Journal of Botany*, *84*(5), 844–851. https://doi.org/10.1139/b06-036

[8] Beltagi, M. S., M. A. I., & F. H. M. (2006). Induced Salt Tolerance in Common Bean (Phaseolas vulgaris L.) by Gamma Irradiation. *Pakistan Journal of Biological Sciences*, *9*(6), 1143–1148. https://doi.org/10.3923/pjbs.2006.1143.1148

[9] Ali, N., Rafiq, R., Zaib-un-Nisa, Wijaya, L., Ahmad, A., & Kaushik, P. (2024). Exogenous citric acid improves growth and yield by concerted modulation of antioxidant defense system in brinjal (Solanum melongena L.) under salt-stress. *Journal of King Saud University - Science*, *36*(1), 103012. https://doi.org/10.1016/j.jksus.2023.103012

[10] Ayub, Q., Masaud Khan, S., Hussain, I., Gurmani, A. R., Naveed, K., Mehmood, A., Ali, S., Ahmad, T., ul Haq, N., & Hussain, A. (2021a). Mitigating the adverse effects of NaCl salinity on pod yield and ionic attributes of okra plants by silicon and gibberellic acid application. *Italus Hortus*, *28*(1), 59. https://doi.org/10.26353/j.itahort/2021.1.5973

[11] Haq, I.-, Khan, A. A., Khan, I. A., & Azmat, M. A. (2012). Comprehensive screening and selection of okra (Abelmoschus esculentus) germplasm for salinity tolerance at the seedling stage and during plant ontogeny. *Journal of Zhejiang University SCIENCE B*, *13*(7), 533–544. https://doi.org/10.1631/jzus.B1200027

[12] Muhammad Adnan Shahid, Muhammad Aslam Pervez, Rashid Mukhtar Balal, Rashid Ahmad, Chaudhary Muhammad Ayyub, Tahira Abbas, & Naheed Akhtar. (2011). Salt stress effects on some morphological and physiological characteristics of okra (Abelmoschus esculentus L.). *Soil Environ.*, *30*(1), 66–73.

[13] Ma, J., Du, G., Li, X., Zhang, C., & Guo, J. (2015a). A major locus controlling malondialdehyde content under water stress is associated with Fusarium crown rot resistance in wheat. *Molecular Genetics and Genomics*, *290*(5), 1955–1962. https://doi.org/10.1007/s00438-015-1053-3

[14] Hnilickova, H., Kraus, K., Vachova, P., & Hnilicka, F. (2021). Salinity Stress Affects Photosynthesis, Malondialdehyde Formation, and Proline Content in Portulaca oleracea L. *Plants*, *10*(5), 845. https://doi.org/10.3390/plants10050845

[15] Yazici, I., Türkan, I., Sekmen, A. H., & Demiral, T. (2007). Salinity tolerance of purslane (Portulaca oleracea L.) is achieved by enhanced antioxidative system, lower level of lipid peroxidation and proline accumulation. *Environmental and Experimental Botany*, *61*(1), 49–57. https://doi.org/10.1016/j.envexpbot.2007.02.010

[16] Chaparzadeh, N., D'Amico, M. L., Khavari-Nejad, R.-A., Izzo, R., & Navari-Izzo, F. (2004). Antioxidative responses of Calendula officinalis under salinity conditions. *Plant Physiology and Biochemistry*, *42*(9), 695–701. https://doi.org/10.1016/j.plaphy.2004.07.001

[17] Mittler, R. (2002). Oxidative stress, antioxidants and stress tolerance. *Trends in Plant Science*, *7*(9), 405–410. https://doi.org/10.1016/S1360-1385(02)02312-9

[18] Azooz, M. M., A.M. Ismail, & M.F.A. Elhamd. (2009). Growth, lipid peroxidation and antioxidant enzyme activities as a selection criterion for the salt tolerance of three maize cultivars grown under salinity stress. *INTERNATIONAL JOURNAL OF AGRICULTURE & BIOLOGY*, *11*(1).

[19] Liang, Y., Sun, W., Zhu, Y.-G., & Christie, P. (2007). Mechanisms of silicon-mediated alleviation of abiotic stresses in higher plants: A review. *Environmental Pollution*, *147*(2), 422–428. https://doi.org/10.1016/j.envpol.2006.06.008